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Cyanobacterial diversity in eight lentic habitats of temples in Southwest India

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ABSTRACT

Lentic water bodies of temples (tanks, ponds and lakes) are traditionally conserved unique heritage ecosystems over a long period and serve as potential sources of groundwater recharge. Unlike other aquatic ecosystems, they have fairly less human interference, hence the water quality and aquatic biodiversity seem to be unique. Among the eight temple lentic habitats, the cyanobacterial population (18 genera in 9 families) was dominated by non-heterocystous forms (17 spp.) followed by unicellular forms (11 spp.) and heterocystous (8 spp.) forms. Among the 36 species, the genus *Phormidium* showed dominance (6 spp.), followed by *Oscillatoria* (5 spp.). *Aphanocapsa biformis* and *Chroococcus montanus* were highly dominant followed by *Oscillatoria curviceps*, *O. subbrevis* and *Chlorogloea fritschii*. None of the species was confined to a single location. The species richness, diversity and equitability were highest in Karinjeshwara temple pond, while they were least in Nanjundeshwara temple tank except for Shannon's diversity. A total of 11 rare species of cyanobacteria were documented in this study (*Anabeana oscillatoroides, A. spiroides, Gleocapsa stigophila, Lyngbya sordida, Microcystis aeruginosa, Oscillatoria limosa, Phormidium ambiguum, Ph. anomala, Ph. foreaui, Planktothrix peronata* and *Scytonema simplex*). The overall water quality reveals its pristine nature and being oligotrophic seems to support many nitrogen-fixing cyanobacteria. Temple lentic ecosystems serve as repositories of a variety of cyanobacteria of nutritional, medicinal, agricultural and industrial significance.

Key words: Aquatic habitats, conservation, cyanophyceae, heritage, species richness, water quality

1. INTRODUCTION

Temple lentic water bodies are freshwater habitats traditionally linked with the temples in India as part of temple structure and heritage (Tadgell, 1990). Usually, almost all such lentic sources are fairly unpolluted due to restricted use. The major water source for these lentic habitats is rainwater, thus in addition to its usefulness to temple activities, they enable rainwater harvesting as well as groundwater recharge (Narchonai et al., 2019). These water bodies support a variety of flora, fauna and microbes as they receive organic and

inorganic materials of routine rituals. Owing to the development of microhabitats and the availability of sunlight, temple lentic water bodies serve as hotspots for various plankton including algae and bacteria. Phytoplankton being primary producers, balance the water quality of lentic habitats by enrichment of nutrients and energy to support other organisms (Oscar et al., 2014; Denise et al., 2018).

In addition to microalgae, temple lentic ecosystems sustain a variety of cyanobacteria (filamentous and unicellular forms) (Muthukumar et al., 2007; Umarani et al., 2017; Narchonai et al., 2019; Rao and Rao, 2020; Jayakumar and Pandiyan, 2021). Cyanobacteria are cosmopolitan and widely distributed in water bodies, irrespective of their geographic locations and physicochemical features (Thajuddin and Subramanian, 2005; Ram & Paul, 2021). According to Liu et al. (2004), the presence of cyanobacteria prevents contamination in water bodies by anthropogenic interference. Cyanobacterial richness, diversity and dynamics are influenced by the physicochemical characteristics of different temple lentic waters in India (Anuja and Chandra, 2012; Bajpai et al., 2013; Girish et al., 2014; Sankaran and Thiruneelagandan, 2015; Palanivel and Umarani, 2016). There are limited reports available on the species richness and diversity of cyanobacteria in temple lentic waters in the Southern Karnataka region. Therefore, the present study was undertaken with the objectives to document the richness and diversity of cyanobacteria in the eight temple lentic bodies of Southern Karnataka along with physicochemical characteristics.

2. MATERIALS AND METHODS

Eight temple lentic sources have been chosen for the survey of cyanobacteria along with physicochemical characteristics during July-August 2021 (Table 1). Four different samples were aseptically collected (morning hours: 9-11 am) from each lentic body (water, floating leaves, floating woody debris and submerged wall/rock scraping). Water samples were filtered through nylon mesh to assess the presence of cyanobacteria. Filtered water scum, scrapings of rinsed substrates (leaves and woody debris) and wall scrapings were transferred into distilled water in Petri plates for observation under stereo (Nikon, Model ECLIPSE Ts2-FL 244472, Nikon Corporation, Tokyo, Japan) and high power (Nikon, Model ECLIPSE Ni-U 941966, Nikon Corporation, Tokyo, Japan) microscopes for identification (Desikachary, 1959; Anagnostidis and Komarek. 1998; Komárek and Anagnostidis, 2005). The presence or absence of species of cyanobacteria in each sample was recorded. The frequency of occurrence (%) [(number of samples possess a specific species/total number of samples assessed) × 100] and relative abundance (%) [(frequency of occurrence of a specific species/total frequency of occurrence) × 100] of cyanobacteria was assessed. Based on the overall species richness and frequency of occurrence of cyanobacteria, the Simpson and Shannon diversities along with Pielou's equitability were calculated for each temple lentic habitat (Pielou 1975, Magurran 1988).

Table 1. Details of eight temple lentic habitats studied for cyanobacteria southwest India.

	Code	Geographic location	*Geographic coordinates*
Karinjeshwara *temple pond*	KP	Bantwala, Dakshina Kannada District, Karnataka	12°54′51.12″N; 75°3′34.56″E
Varanga Neminath Basadi lake	VL	Hebri, Udupi District, Karnataka	13°23′46″N; 75°00′15″E
Talakaveri temple pond	TP	Bhagamandala, Kodagu District, Karnataka	12°22′48″N; 75°31′12″E
Devaramane Kalabhairaveshwara temple pond	DP	Mudigere, Chikkamagalore District, Karnataka	13°03′41.7″N; 75°32′19.5″E
Varadamoola temple pond	VP	Varadahalli, Shimoga District, Karntaka	14°7′32″N; 74°59′15″E
Halebeedu temple tank (Dwarasamudra lake)	HT	Halebeedu, Hassan District, Karnataka	13°12′56.52″N; 75°59′29.04″E
Nanjundeshwara temple tank	NT	Nanjanagudu, Mysore District, Karnataka	12°7′8″N; 76°41′33″E
Ranganathaswamy *temple* tank	RT	Srirangapatna, Mandya District, Karnataka	12°25′29.11″N; 76°40′47″E

Additional four water samples were collected from four corners of each water body for physicochemical analysis. Temperature (mercury thermometer), pH, conductivity and total dissolved solids were assessed on the sampling site by the Water Analyzer 371

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(Systronics, Gujarat, India). Total alkalinity, dissolved oxygen (fixed water samples), biochemical oxygen demand, dissolved organic matter, free carbon dioxide, total hardness, nitrate, phosphate, sulfate, chloride and silicate were assessed based on the APHA (1998).

3. RESULTS AND DISCUSSION

Thirty-six cyanobacterial species (18 genera and 9 families) consisting of non-heterocystous forms (17 spp.), unicellular forms (11 spp.) and heterocystous forms (8 spp.) were recorded in eight temple lentic waters (Figure 1). Among the genera, *Phormidium* was dominant (6 spp.) followed by *Oscillatoria* (5 spp.) and *Anabeana* and *Chroococcus* (each with 3 spp.) (Table 2). The frequency of occurrence of *Aphanocapsa biformis* and *Chroococcus montanus* was highest followed by *Oscillatoria curviceps*, *O. subbrevis*, *Chlorogloea fritschii* and *Chroococcus minor*. None of the species were confined to specific temple lentic bodies. The species richness, diversity and equitability were highest in Karinjeshwara temple pond, while they were lowest in the Nanjundeshwara temple tank except for Shannon's diversity (Table 3). Eleven rare species of cyanobacteria documented in this study include *Anabeana oscillatoroides, A. spiroides, Gleocapsa stigophila, Lyngbya sordida, Microcystis aeruginosa, Oscillatoria limosa, Phormidium ambiguum, Ph. anomala, Ph. foreaui, Planktothrix perornata* and *Scytonema simplex*. Representative species of cyanobacteria found in this study are presented in Figure 2.

Table 2. Cyanobacterial population, frequency of occurrence (FO) and relative abundance (RA) in eight temple lentic habitats (n=32) (*, see Table 1 for habitat code; **, rare species).

	Temple lentic habitats*								FO (%)	RA (%)
	KP	VL	TP	DP	VP	HT	NT	RT		
Non-heterocystous forms										
Oscillatoria curviceps C. Agardh ex Gomont	+	+	+	+	+	+	+	+	87.5	5.0
Oscillatoria subbrevis Schmidle	+	+	+	+	+	+	+	+	79.2	4.5
Phormidium chlorinum Umezaki & Watanabe	+	+	+	+	+	+	+	-	66.7	3.8
Phormidium ornatum Anagnostidis & Komarek	+	+	+	+	+	+	+	-	62.5	3.6
Leptolyngbya fragilis (Gomont) Anagnostidis & Komarek	+	+	+	-	+	+	-	+	58.3	3.3
Oscillatoria earlei N.L. Gardner	+	+	+	+	+	+	+	-	58.3	3.3
Porphyrosiphon dendrobius (Bruhl & Biswas) Anagnostidis & Komarek	+	-	+	-	+	+	-	+	54.2	3.1
Leptolyngbya boryana (Gomont) Anagnostidis & Komarek	+	-	-	+	-	+	+	+	45.8	2.6
Oscillatoria amoena Gomont	+	+	-	-	+	+	-	+	45.8	2.6
Phormidium versicolour Wartmann ex Gomont	+	+	+	-	+	+	-	+	45.8	2.6
***Lyngbya sordida* Gomont	-	-	-	-	-	+	+	+	33.3	1.9
***Phormidium anomala* Rao C.B.	+	-	-	-	-	+	-	+	33.3	1.9
***Phormidium foreaui* (Fremy) Umezaki & Watanabe	+	-	+	-	-	-	+	-	33.3	1.9
***Phormidium ambiguum* Gomont	+	+	-	-	-	-	-	+	29.2	1.7
***Planktothrix perornata* (Skuja) Anagnostidia & Komarek	-	+	-	+	+	-	-	-	29.2	1.7
***Oscillatoria limosa* C. Agardh ex Gomont	+	-	+	-	-	-	-	-	25.0	1.4
Spirulina princeps West & West G.S.	-	-	+	-	-	+	-	-	25.0	1.4
Total species (17)	**14**	**10**	**11**	**7**	**10**	**13**	**8**	**10**		
Unicellular forms										
Aphanocapsa biformis A. Braun	+	+	+	+	+	+	+	+	100	5.7
Chroococcus montanus Hansgirg	+	+	+	+	+	+	+	+	100	5.7
Chlorogloea fritschii Mitra	+	+	+	+	+	+	-	+	75.0	4.3
Chroococcus minor Nageli	+	-	+	+	+	+	+	+	70.8	4.0
Synnechocystis pevalekii Erceg.	+	+	+	+	+	+	+	+	66.7	3.8
Chroococcus turgidus Nageli	+	+	+	+	-	+	+	+	58.3	3.3
Aphanocapsa littoralis Hansg.	+	+	+	+	-	+	-	+	54.2	3.1
Gloeocapsa gelatinosa Kützing	+	-	+	+	+	+	-	+	45.8	2.6
Merismopedia tenuissima Lemmermann	+	+	-	+	-	+	-	+	45.8	2.6
***Gleocapsa stigophila* Rabenh	+	-	+	-	-	-	-	+	29.2	1.7
***Microcystis aeruginosa* (Kützing) Kützing	+	-	-	-	-	-	+	-	16.7	1.0
Total species (11)	**11**	**7**	**9**	**9**	**6**	**9**	**6**	**10**		
Heterocystous forms										

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Trichormus arvensis (Rao C.B.) Komarek & Anagnostidis	+	+	-	+	+	+	-	+	62.5	3.6
Nostoc ellipsosporum Rabenh.	+	-	-	+	+	+	+	+	50.0	2.8
Anabaena fertilissima Rao C.B.	+	+	+	-	+	+	-	+	41.7	2.4
Nostoc commune Vaucher ex Bornet & Flahault	+	+	+	+	+	+	-	+	41.7	2.4
***Anabaena spiroides* Klebahm	+	-	+	-	-	+	-	-	29.2	1.7
Anabaena variabilis Fritsch	-	+	+	+	-	+	+	-	29.2	1.7
***Anabeana oscillatoroides* Bharadwaja	+	-	-	-	-	+	-	-	16.7	1.0
***Scytonema simplex* Bharadwaja	-	-	-	+	+	-	-	-	16.7	1.0
Total species (8)	**6**	**4**	**4**	**5**	**5**	**7**	**2**	**4**		

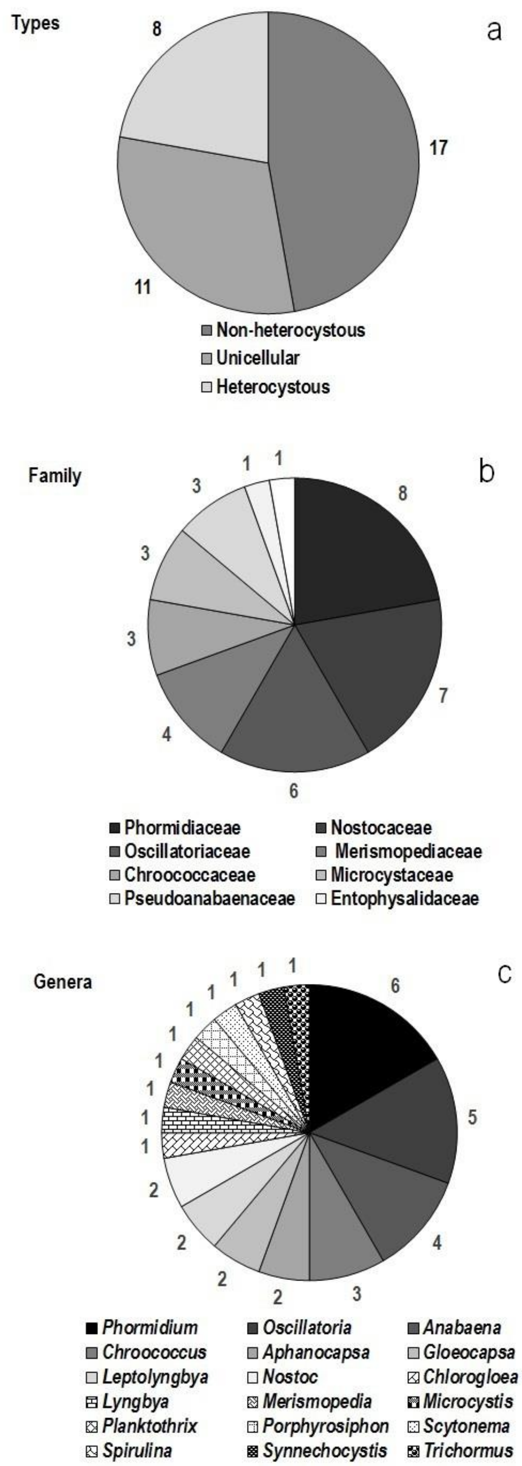

Figure 1. Distribution of different types, family and genera of cyanobacteria in temple lotic habitats.

Table 3. Species richness and diversity of cyanobacteria in the eight temple lentic habitats (*, see Table 1 for habitat code).

	Temple lentic habitats*							
	KP	VL	TP	DP	VP	HT	NT	RT
Species richness	31	21	24	21	21	29	16	24
Simpson's diversity	0.987	0.735	0.926	0.885	0.775	0.885	0.645	0.684
Shannon's diversity	1.652	1.055	1.365	1.119	1.095	1.358	1.215	1.274
Pielou's equitability	0.782	0.545	0.715	0.652	0.622	0.628	0.594	0.616

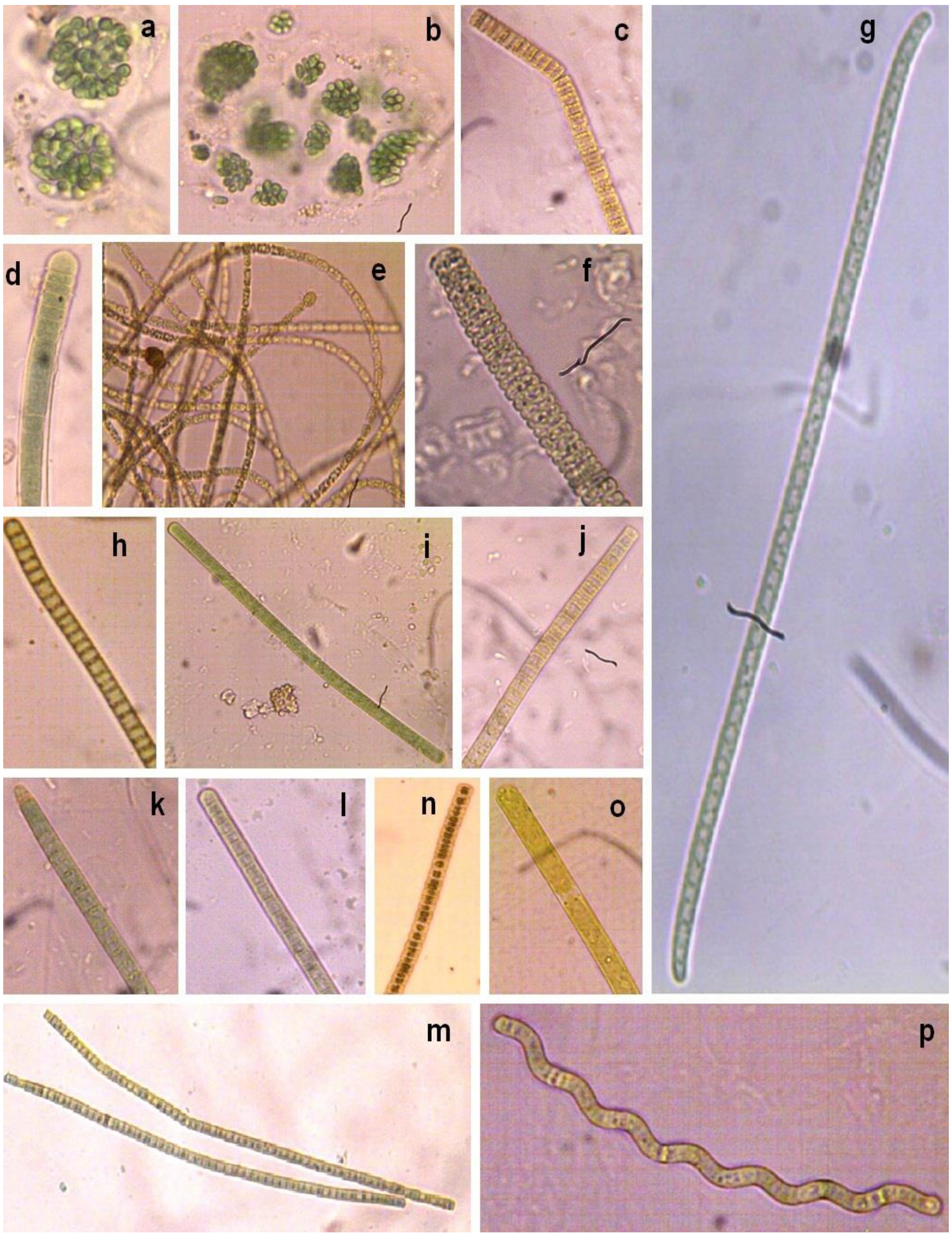

Figure 2. Selected species of cyanobacteria: *Aphanocapsa littoralis* (a), *Chlorogloea fritschii* (b), *Leptolyngbya fragilis* (c), *Lyngbya sordida* (d), *Nostoc ellipsosporum* (e), *Oscillatoria amoena* (f), *Oscillatoria curviceps* (g), *Oscillatoria earlei* (h), *Oscillatoria limosa* (i), *Oscillatoria subbrevis* (j), *Phormidium anomala* (k), *Phormidium chlorinum* (l), *Phormidium foreaui* (m), *Planktothrix perornata* (n), *Porphyrosiphon dendrobius* (o) and *Spirulina princeps* (p).

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Cyanobacterial population in the present study is closer to other lentic bodies such as Dhenupuriswara and Thiyagarajaswamy temples in Tamil Nadu (36 vs. 38 spp.), while higher than Nageswara, Pandakkal and Palloor temples (36 vs. 17-29 spp.) (Girish et al., 2014; Palanivel and Umarani, 2016; Umarani et al., 2017). Dominance of *Oscillatoria curviceps, O. subbrevis* is similar to the findings in temple tanks of Kerala as well as Orissa (Swain et al., 1993; Maya et al., 2000). *Microcystis aeruginosa* in temple lentic bodies was found in two sampling locations (Karinjeshwara temple pond and Nanjundeshwara temple tank) with low relative abundance (1.4%). It is known for toxic blooms in different water bodies (Mohan et al., 2020; Vanderley et al., 2021). Swain et al. (1993) recorded the bloom of *M. aeruginosa* in two temple ponds at Puri (India) owing to the high content of nitrogen and phosphorous.

Based on the extent of 15 water parameters, almost all the lentic bodies studied fall under the oligotrophic category (Table 4). As many as 13 parameters were the lowest in the Talakavari temple pond, while only two parameters were minimum in the Devaramane temple pond. Maximum values for six, four, three and two parameters in lentic bodies of the Ranganathaswamy, Varanga Neminath Basadi, Nanjundeshwara and Halebeedu temples, respectively. Interestingly, the Karinjeshwara temple pond with almost the median values of 15 water parameters showed the highest species richness, diversity and equitability of cyanobacteria (see Table 3). With three highest water parameters in the Nanjundeshwara temple tank showed the least cyanobacterial species richness, Simpson's diversity and equitability. However, the lake Varanga Neminath Basadi with the four highest water parameters showed the lowest Shannon's diversity.

Table 4. The physicochemical properties of water samples of eight temple lentic habitats (n=4, mean; *, see Table 1 for habitat code).

	Temple lentic habitats*								Range
	KP	VL	TP	DP	VP	HT	NT	RT	
Temperature (°C)	27.0	26.5	28.6	26.5	27.0	28.0	29.0	27.2	26.5–29
pH	8.4	8.5	7.9	8.0	8.0	8.2	8.6	8.5	7.9–8.6
Conductivity (μS/cm)	250.7	232.6	120.0	212.0	231.0	223.3	245.6	282.3	120–282.3
Total alkalinity (mg/l)	155.5	188.6	65.2	85.5	95.3	112.8	130.3	160.6	65.2–188.6
Dissolved oxygen (mg/l)	7.9	8.0	8.9	8.6	8.7	8.0	7.8	7.8	7.8–8.9
Biochemical oxygen demand (mg/l)	14.2	20.2	8.4	9.0	10.7	14.4	16.8	15.3	8.4–20.2
Dissolved organic matter (mg/l)	5.2	7.2	3.6	4.8	5.4	6.6	7.5	7.8	3.6–7.8
Total dissolved solids (mg/l)	235.0	255.0	182.0	148.3	220.0	216.6	232.3	275.0	148.3–275
Free carbon dioxide (mg/l)	20.5	25.1	10.7	15.3	12.6	25.4	24.1	18.6	10.7–25.4
Total hardness (mg/l)	110.6	180.6	85.6	92.5	138.5	155.0	125.0	235.6	85.6–235.6
Nitrate (mg/l)	8.1	10.3	5.2	7.6	6.6	7.2	8.7	7.7	5.2–10.3
Phosphate (mg/l)	12.5	18.4	6.3	10.7	12.0	20.3	19.2	18.3	6.3–20.3
Sulfate (mg/l)	4.3	6.3	3.2	4.9	4.8	5.7	5.3	6.8	3.2–6.8
Chloride (mg/l)	30.8	32.8	18.2	20.5	25.8	28.4	30.2	26.8	18.2–32.8
Silicate (mg/l)	12.3	16.8	10.5	12.8	10.7	20.6	25.7	22.8	10.5–25.7

Many cyanobacteria fix atmospheric nitrogen and help to maintain a close balance between nitrogen and phosphate limitations (Karl et al., 2002). Nitrate content in all lentic bodies studied was lower than phosphate (ratio, <1) is a glaring observation. Possibly with low nitrate content, these lentic bodies consist of eight nitrogen-fixing heterocystous forms (range, 2-7 spp.) that seem to compensate for the nitrate deficiency.

Many parameters studied in the current study confirm the WHO (1989) stipulated water standards. The alkaline pH value of temple lentic bodies ranged between 7.9 and 8.6, which is almost close to the WHO standard (6.5–8.5). The electrical conductivity depicts the level of pollution and purity of water bodies (Shrestha et al., 2017). Its measurements agree with the WHO standard (250 μS/cm) except for the Ranganathaswamy temple tank (282.3 μS/cm). The total dissolved solids indicate a load of dissolved substances, which varied between 130 and 280 ppm in temple lentic bodies are within an acceptable range of WHO. The extent of total hardness of temple lentic waters ranged from 85.6-235.6 mg/l, it is within the WHO recommended standard (600 mg/l). Likewise, the nitrate content ranged between 5.2 and 10.3 mg/l, which is within the WHO recommended standard (50 mg/l). Phosphorous plays an important role in biochemical processes and is a major factor in the eutrophication of surface waters (Oladeji et al., 2016). In our study, the phosphate content ranged between 6.3 and 20.3 mg/l, it is greater than the WHO standard. The sulfate content ranged between 3.2 and 6.8 mg/l, it is within the WHO recommended concentration (500 mg/l). The chloride level recorded was ranged from 18.2-32.8 mg/l, which is below the WHO recommended concentration (250 mg/l).

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Many cyanobacteria have biotechnological significance (e.g. nutrition, health and agriculture) (Tajuddin and Subramanian, 2005). For example, *Spirulina princeps* are used widely in food products (https://patents.google.com/patent/CN1473509A/en). It was found in two locations in our study (Talakaveri temple pond and Halebeedu temple tank). Similarly, *Nostoc commune* and *N. ellipsosporum* possess nutritional value (Chu and Tsang, 1988; Liao et al., 2015), while cryptophycin produced by *N. commune* has antiviral and anticancer potential (Knubel et al., 1990; Smith et al, 1994). These species were recorded in seven and six lentic waters, respectively. Mucilaginous polymers produced by cyanobacteria helps in moisture retention, soil binding and enhancement of humus content in soil (Maqubela et al., 2009; Prasanna et al., 2013; Singh et al., 2014). Cyanobacteria are also known for the production of amino acids and indole-3-acetic acid by intimate association with roots of wheat (Hussain and Hasnain, 2011). Many heterocystous forms produce pigments (e.g., phycocyanin and phycoerythrin) (Rodriguez et al., 1989; Simeunovic et al., 2013). These pigments have various applications in food, pharmaceutical and cosmeceutical industries as natural coloring agents (Patterson, 1996). Phycocyanin is also used in immunodiagnostics as well as immunomodulation in cancer therapy (Benedetti et al., 2004). Besides nitrogen fixation, eight heterocystous cyanobacteria found in our study are valuable resources for several industrial applications.

4. CONCLUSION

Temple lentic ecosystems are a valuable source of flora, fauna and microbes. Owing to fairly less anthropogenic interference, these water bodies sustain or serve as repositories of many cyanobacteria of nutritional (proteins, vitamins, lipids and colors), health (enzymes, pharmaceuticals and toxins) and agricultural (feed, fertilizer and soil conditioner) significance. The oligotrophic feature of temple lentic habitats studied supports a fair population of heterocystous cyanobacteria of agricultural importance in nitrogen fixation to enrich the soil and waters. Some of the strains of cyanobacteria are also of commercial significance especially in the production of bioenergy and biofuel (hydrogen and methane). Cyanobacteria serve as an inexpensive source of biomass that could be generated by harnessing the sunlight by their simple structure (unicellular, multicellular, filamentous and heterocystous forms), rapid growth and ease of harvest. Future studies need to consider the heritage lentic ecosystems for useful cyanobacterial strains in biotechnological innovations.

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Ethical approval

The ethical guidelines are followed in the study for the collection of samples and studied with the help of Dr. Mahadevakumar, Karnataka State Open University, Mysore, India & Department of Biosciences, Mangalore University, India.

Authors' contributions

All authors contributed equally.

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Conflicts of interests

The authors declare that there are no conflicts of interests.

Data and materials availability

All data associated with this study are present in the paper.

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